Appendix 1

Comments from Joel Hoffman, U.S. EPA

Comments on proposal "SAMPLING AND ANALYSIS PLAN WITH QUALITY ASSURANCE PROJECT PLAN FOR THE FRANK HOLTEN STATE PARK LAKES", submitted to US EPA Region 5 by Veolia ES Technical Solutions, LLC

Review provided at the request of Todd Ramalay, Environmental Scientist, RCRA Programs Section, U.S. EPA - Region 5.

Summary: The general goal of this project is: "to test the fish, water, and sediment in the Frank Holten State Park Lakes." The park includes three lakes: Whispering ("Lake 1"), Willow ("Lake 2"), and Grand Marais ("Lake 3") lakes. Whispering and Willow lakes appear to share a direct surface water connection. The combined area and shoreline distance of Whispering and Willow lakes are similar to those of Grand Marais Lake. The lakes are described as shallow and supportive of a typical cool-warm water fish community, including common carp (*Cyprinus carpio*); channel catfish (*Ictalurus punctatus*); various centrarchids, including largemouth bass (*Micropterus salmoides*), white crappie (*Pomoxis annularis*), black crappie (*Pomoxis nigromaculatus*), and bluegill (*Lepomis macrochirus*); as well as muskellunge (*Esox masquinongy*). The authors propose to sample sediment cores, water, and fish for mercury concentrations, as well as fish tissue for the nitrogen stable isotope ($\delta^{15}N$) abundance to determine trophic level. From each lake, two sediment cores (one from the center of the lake and one from the littoral zone), three water samples, and four fish species (three individuals of each species from two size-classes) will be sampled. The output of the study is not described, but is presumably a report detailing the mercury concentrations found in the samples, as well as an estimate of the trophic level of the fish.

Review: In general, the study has two substantial flaws that need to be addressed. My first concern with the study is the general lack of a scientific study design. The origin of the problem is the lack of a specific objective. For example, if the goal is to compare the three lakes, then what amount of difference between lakes would be considered important? How many samples (given typical variability in the environment) would be required to detect such a difference? Alternatively, if the goal is determine the mercury concentration of fish most likely to be consumed from the lake, are data available to determine the most popular species and sizes chosen for consumption? It is not clear that the species or sizes chosen are the ones most relevant to evaluate human health risks. Without a clear statement of objectives, it is impossible to evaluate the critical aspects of the study design – numbers of samples and sample allocation.

My second concern with the study is that it will produce insufficient data to utilize the nitrogen stable isotope values (δ^{15} N) to estimate trophic levels for the fish species sampled. The δ^{15} N value in fish tissue is useful for estimating trophic level because the $\delta^{15}N$ value in consumers reflects that of their prey. In aquatic organisms, the δ^{15} N value measured increases about 3-4‰ with each trophic level. This is called the trophic fractionation. The key element is that the $\delta^{15}N$ value in the fish is derived from the fish's prey and, in turn, the fish's prey δ^{15} N is determined by its diet (and so on through the food web). Thus, without information on the prey δ^{15} N, the trophic level cannot be determined. The underlying δ^{15} N signature of the prey is often referred to as a $\delta^{15}N$ baseline (or base). The equation for determining trophic level (TL) based on $\delta^{15}N$ is: TL = λ + ($\delta^{15}N_{consumer} - \delta^{15}N_{base}$) / Δ_n , where λ is the trophic position of the plant or organism used to determine $\delta^{15}N_{base}$, $\delta^{15}N_{base}$ is usually estimated from either the particulate nitrogen or a representative grazing invertebrate, and Δ_0 is the average trophic fractionation (typically 3-4%; for review, see Post, 2002, Ecology 83:703-718). In lakes, however, the littoral, benthic, and pelagic food webs rely on different primary producers (littoral on epiphytic periphyton and benthic microalgae, benthic on degraded algae that sinks to the bottom, and pelagic on phytoplankton). These different primary producers often have different δ^{15} N values due to differences in the local nitrogen cycling. If the primary producers have different $\delta^{15}N$ values, then there will be different $\delta^{15}N$ baselines for the littoral, benthic, and pelagic food webs. The effect of this variation between food webs is that a top-level pelagic predator

will have a different $\delta^{15}N$ value than a top-level littoral predator. To estimate trophic level for the fishes identified, the authors will have to also measure $\delta^{15}N$ values in pelagic, littoral and benthic grazers (i.e., zooplankton, amphipods or snails, and chironomids), as well as common fish prey species. These data are necessary to determine the $\delta^{15}N$ base and correctly estimate a trophic level in the case where a fish species is feeding on prey from multiple food webs (e.g., littoral and benthic prey items).

Below are a few minor comments. At this time, I chose not to provide thorough comments on the methodology given the concerns identified above.

Regarding field methods, the general collection methods and techniques are appropriate. The authors propose to use EPA methods where applicable. The Field Record Form for Fish should include a wet weight, as stable isotope values are most appropriately expressed as a function of weight (somatic growth) rather than length. The water sample form should indicate a sample depth – it is not clear if they are taking samples from the epilimnion, metalimnion, or hypolimnion. For the cores, a bottom depth should also be recorded. Also, because pH is an important parameter for mercury accumulation, pH of the lake water should be measured.

Regarding lab methods, the general collection methods and techniques are appropriate. As before, the authors propose to use EPA methods where applicable.

Joel Hoffman, PhD Research Biologist

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Appendix 2

Comments from Christopher Knightes, U.S. EPA

Chris and Todd,

I've gone through the Lake Study that you sent me. I really just focused on the sampling strategy. I can't really speak to the analytical approach, as I'm not out in the field doing the actual sampling.

I'm not very clear on what the point of the N-isotope study is for. I understand that they can use it to determine trophic status, and I'm supposing they're hoping to argue that it'll be less than 4? From talking to Craig Barber (our fish bioaccumulation expert), he believes that Carp will fall in around trophic level 2.5, and the other 3 will fall somewhere between 3 and 4. That being said, what would be more useful would be to calculate a site-specific BAF using observed MeHg concentrations and fish tissue concentrations.

What is the goal of sampling the smaller fish? Both Craig and I agree that sampling for 3 fish isn't going to give you enough information, I'd recommend getting at least 5 of each time, hiking up the total number of samples for each fish species to 10. There is going to be a lot of scatter with these samples, so having more will give a better representation of the variance and the relationship between Hg fish tissue and age, length, weight. I'm assuming each fish sample will have a length and weight recorded, age is a little more challenging, but length and weight would work. Craig also suggested that for the "small" vs "large" they should include a factor of at least an order of magnitude difference in weight, but preferably 30x. If a 100g small fish is collected, then a 3000g large fish should be collected.

For the water, 3 isn't going to get very far. I'd recommend doing 9, and particularly doing it at different times. Feasibly every 2 weeks during the growing season. Again, these MeHg concentrations in water are going to be bouncing all over the place. To get a solid grasp on the BAFs, you're going to need to have a solid representation on the variance and mean of the MeHg. You could also do 5 times and take 2 samples at different places in the lake. There will be spatial variability as well in water, but the temporal variability will be more pronounced. I know it sounds like the MeHg sampling is a lot, *but* to really nail down the BAFs, without ending up with a log BAF of 6 +/-1.5, they'll likely need it.

I saw that they're doing filtered and unfiltered. I believe that the national BAFs use unfiltered MeHg (because filtered is more costly), but filtered MeHg would be better from a scientific standpoint, but the calculated BAF won't reflect the national BAF.

What is the point of the sediment cores? These would give a gage on the %MeHg in the sediments, but sediment mercury isn't the best correlation to fish tissue concentrations. If sediment cores are suggested, it would be much more productive to have multiple cores across the lake. I would suggest 5. The issue here is sediments are incredibly variable. If you go 5' in one direction, you could easily see concentrations go up or down an order of magnitude. Since the mean is what you're really interested in, getting several to get a good representation would be critical. Doing a core will give a nice representation the history of Hg accumulation in the sediments, but I'm not sure what the point is. Are they trying to make an argument that they aren't responsible for increased Hg? If so, I'd have trouble using a single core to tell me that, again, the numbers are going to be all over the place as sediments are so heterogeneous.

With respect to the 3 lakes, looking at the 3 with different characteristics, I wouldn't assume that they're behaving the same. It would not surprise me to have %MeHg, BAFs and fish tissue Hg concentrations to be different in the 3 different lakes/ponds.

Is the receptor for risk focused on human health or wildlife health as well? I was wondering why the collection of the smaller fish. I'm guessing the small fish would be whole body and the larger would be fillet? I wonder this because aren't small fish hard to get fillets from? If they're done differently, than a conversion factor will need to be done to get them on the same page. So it'd be easier if they were both the same.

Let me know if this helps or if you have any other questions.

Chris

PS Of course, you can always dive into the process model route, but I think you may be able to avoid digging into the weeds with modeling by doing a clean sampling approach, coming up with the %MeHg for the lake (with variance) and BAFs (with variance).

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Appendix 3

Comments of Thomas Hornshaw

MEMORANDUM

DATE:	September 2.	2011
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TO: Todd Ramaly

CC: Ted Dragovich

FROM: Tom Hornshaw

SUBJECT: Veolia Fish Mercury Samples

I have reviewed the Sampling and Analysis Plan with Quality Assurance Project Plan for the Frank Holten State Park Lakes as requested. This document will, when modified by the suggested changes below, provide an acceptable snapshot of the current status of methylmercury in the edible portions of fish, and a basis for comparison with historical data on mercury levels in fish from the Holten Lakes. However, as we discussed in our phone conversation, there is concern for predicting the levels of mercury in the edible portions of various fish species in the event that the Veolia incinerator emits mercury at the maximum amount, and this study will not generate such information. Thus, it appears that site-specific estimates of the conversion of inorganic mercury to methylmercury in water and sediments and site-specific bioaccumulation factors would be needed to forecast what levels of methylmercury might result in the edible portions of Holton Lakes fish due to operation of the incinerator at maximum mercury output.

Suggested changes include:

- p. 9 It is proposed to collect duplicate samples at a rate of 5%, but it is customary to collect duplicates at a rate of 10%.
- p. 16 It is proposed to analyze filet samples of all fish but it is not stated whether these will be skin-on or skin-off filets; to be comparable to the procedures used by the Illinois Fish

- Contaminant Monitoring Program, skin-on scaled filets are preferred for the bass, crappie, and carp samples and skin-off filets for the channel catfish.
- Table 5-2 It is proposed to collect 3 larger and 3 smaller samples of each of the four species to be sampled, but larger and smaller are not further defined. Since the length minimum for largemouth bass is 14 inches this size should be appropriate for the smaller bass samples, and historical data shows that bass in the 16-18 inch range should be available as the larger samples. Historical data also suggest that samples as large as 20 inches should be available for carp and channel catfish and as large as 11 inches for crappies, so these sizes could be used as the larger samples; professional judgment could be used to determine what would be an appropriate size (that would likely be a younger year class) for the smaller samples.
- Detection limits There is no discussion of the anticipated detection limits for water, sediments, and fish in this document, such discussion should be included to give some measure of whether the results will be appropriate to answer the study's questions.

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